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RESPONSE OF YELLOW NUTSEDGE, BARLEY, LETTUCE, SOYBEAN, LITTLE BLUESTEM, CANADA BLUEGRASS, AND CULTIVARS OF TALL FESCUE, RED FESCUE, KENTUCKY BLUEGRASS, AND PERENNIAL RYEGRASS TO EXCESSIVE SEWAGE-SLUDGE APPLIED SOIL ZINC IN AN ACIDIC SOIL.

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#### **SUMMARY**

High soil Zn is a potential limiting phytotoxic heavy metal in many dredged materials and other polluted soils. As soil pH falls, Zn uptake increases and potential for phytotoxicity increases. Copper is usually much less important in potential phytotoxicity of organic matter rich dredged materials and soils because Cu is adsorbed or chelated by the added organic matter. Although it is expected that analysis of dredged materials will eventually provide sufficient information to predict proper management of dredged materials, presently only bioassays of questionable materials can provide that information.

Yellow nutsedge (Cyperus esculentus L) has been used by the Environmental Laboratory, Waterways Experiment Station, US Army Engineers, to bioassay the potential metal phytotoxicity and metal uptake plants grown on dredged materials. Previous research on metal uptake and tolerance by plant species did not include study of Cyperus species. Thus, it was considered useful to compare the relative tolerance of Cyperus vs. other species to metals applied in sewage sludge.

Ten plant species (Cyperus, red fescue, Kentucky bluegrass, tall fescue,

perennial ryegrass, Canada bluegrass, little bluestem, Romaine lettuce, soybean, and barley) were grown in the greenhouse in pots containing Sassafras sandy loam soil amended with 0, 2, 4, or 8% addition of a highly stabilized, strongly acidic, Znrich (43,500 mg Zn/kg dry weight) sewage sludge. The sludge levels provided 0, 870, 1740, and 3480 mg Zn/kg dry soil. The sludge contained only 11 mg Cd/kg, 23 mg Ni/kg, and 700 mg Cu/kg, and made only small changes in these metals in the soil-sludge mixtures. The soil and sludge were selected such that soil-sludge mixtures were pH 5.3 to 5.4 initially. Metals are much more available at low soil pH, and assessment of potential phytotoxicity must consider the "worst case" strongly acidic soil (pH at or below 5.5). Chemical fertilizers were added to allow normal plant growth on the unamended control soil. The soil was handled moist rather than air-dried to preserve soil microbes and promote more normal soil chemistry. For 4 grass species often used in revegetation, 6-9 cultivars were grown to identify which might be more tolerant of excessive soil Zn. Perennial species were harvested after the initial growth, and allowed to regrow for a second clipping to better characterize metal tolerance. Shoots were analyzed to determine if crop or cultivar differences in metal uptake were related to crop or cultivar differences in metal tolerance and to evaluate relative metal uptake among these crops.

Overall, only the known metal tolerant/resistant red fescue cultivar 'Merlin' (obtained from Zn toxic mine spoils in the United Kingdom) grew well at the 1740 and 3480 mg Zn/kg rates. Yields of 'Merlin' were actually higher at the highest Zn rate than the control. Generally, all other red fescue, all bluegrass, and all perennial ryegrass cultivars, Canada bluegrass, little bluestem, and Cyperus suffered severe yield reduction and chlorosis at 1740 and 3480 mg Zn/kg. The dicotyledonous species lettuce and soybean were severely yield reduced and chlorotic with all Zn additions. Barley and several tall fescue cultivars (notably not Kentucky-31, the cultivar widely used in revegetation programs) performed well on 870 and 1740 mg Zn/kg, but suffered phytotoxicity at the 3480 mg Zn/kg rate. In contrast with earlier studies which used ZnSO4 or air-dried soils, increased soil Zn had little effect on plant Mn. In previous studies in this laboratory, Zn-salts additions to air-dried soils caused large increase in plant Mn to the point of Mn phytotoxicity of soybean. With the sludge-Zn and non-dried soil, added Zn had levels. 'Merlin' excluded Zn compared to all other red fescue cultivars, as well as other species tested.

All plants from the sludge-Zn treated soils had increased foliar Zn. 'Merlin' red fescue contained the lowest shoot Zn levels, not exceeding the 500 mg Zn/kg leaves considered indicative of phytotoxicity in most plant species. Lettuce contained the highest Zn levels on the sludge-Zn amended soils, over 3000 mg/kg, with yield reduction averaging 99%. Soybean averaged over 870 mg Zn/kg on the sludge-Zn amended treatments, with 90% yield reduction. Although red fescue, Cyperus, Kentucky bluegrass, Canada bluegrass, soybean, and lettuce generally followed a similar pattern of yield reduction vs. foliar Zn, tall fescue and barley contained much higher foliar Zn at comparable yield reductions. These data indicate that tall fescue and barley tolerate shoot Zn better than the other species.

In contrast with tall fescue and barley, 'Merlin' red fescue tolerates high soil Zn by excluding Zn from the plants.

Cyperus grew well at 870 mg Zn/kg soil (280 mg Zn/kg first clipping shoots), but was severely chlorotic and stunted at 1740 and 3480 mg/kg (contained 720 and 960 mg Zn/kg shoots, respectively). Foliar Zn reached phytotoxic levels in all crops except 'Merlin' red fescue, commonly exceeding 1000 mg/kg on the 4 and 8% sludge rates.

Six crops were harvested a second time after regrowth of perennial species. In all cases except 'Merlin' red fescue, both chlorosis score and foliar Zn concentration were somewhat increased, and normalized yield somewhat decreased on the 4 and 8% sludge treatments compared to the first clipping. Perennial ryegrass and red fescue cultivars and the Canada bluegrass on the higher sludge rates were nearly dead by the end of this growth period. Kentucky bluegrass cultivars did poorly at high rates, while the new turf-type tall fescue cultivars (Houndog; Rebel; Falcon) did fairly well. These cultivars are candidates for field trials on Zn-rich acidic soils. The Kentucky-31 cultivar of tall fescue, which has often been used in revegetation programs in the US, was the least successful tall fescue.

Overall, this research indicates that Cyperus serves as a valid bloassay plant for Zn uptake and tolerance. Cyperus has a much lower slope of plant Zn: soil Zn than do lettuce, soybean, and many of the grass cultivars. However, Cyperus did suffer Zn phytotoxicity when sufficient sludge-Zn was added and soil pH was strongly acidic. Cyperus was not especially tolerant of soil Zn compared to barley and 'Merlin' red fescue. Further, Cyperus had only average tolerance of shoot Zn, suffering significant yield reduction when plant Zn exceeded 400-500 mg/kg. The sludge used did not allow a useful evaluation of relative Cd, Ni, or Pb uptake by these crops. Changes in these elements concentrations in plant shoots likely resulted from phytotoxicity due to the applied Zn rather than changes in metal bioavailability in the soil.

### INTRODUCTION

The Environmental Laboratory of the Waterways Experiment Station, U.S. Army Engineers, has developed an effective bioassay for phytotoxicity and potential food-chain transfer of heavy metals from dredged materials placed in wetland or upland management situations (Folsom and Lee, 1981; Folsom et al., 1981; Lee et al., 1982). This bioassay uses yellow nutsedge (plant species Cyperus esculentus L., hereafter called Cyperus), a weed species which naturally occurs in both upland and flooded soils. Although Folsom and co-workers have characterized the response of Cyperus to many dredged materials and some control cropland soils, an advisory group for the Dredged Material Research Program recommended that the bioassay response of Cyperus could be more useful if it were compared to other plant species in several soil media. Comparisons of Cyperus with other wetland species and some vegetable crops was conducted on dredged materials in the Netherlands by Driel et al. (1985), while the relative responses of Cyperus and several other species was evaluated on metal mine spoil by B.E. Davies (unpublished).

The present report describes another experimental effort to evaluate the metal uptake and tolerance of Cyperus compared to other species which have been studied frequently in research on potential risks of trace elements in land-applied sewage sludge. Because lettuce, soybean, and other species have been used in many studies of the effects of sludge-applied trace elements, the advisory group felt that comparison of Cyperus with representative crops used in sludge metal risk evaluation, on sludge-amended soils, would provide further useful information to calibrate the responses of Cyperus and assist the Army in interpreting results from bioassays. Although the original plan of this research was to do the comparison with aged sludge from Baltimore, MD (rich in Zn, Cu, and Ni) as used by Chaney et al. (1978), this sludge was no longer available. The final experiments were conducted with an aged sludge high in Zn, but with normal levels of the other elements. High soil Zn is a potential limiting phytotoxic heavy metal in many dredged materials and other polluted soils. As soil pH falls, Zn uptake increases and the potential for Zn phytotoxicity is more severe (Chaney et al., 1975; White et al., 1979a; 1979c; Francis et al., 1985). Copper is usually much less important in the potential phytotoxicity of organic matter rich dredged materials. nusearch summarized by Webber et al. (1981) indicates that sewage sludge Cu does not become phytotoxic at any sludge application rate until the sludge Cu concentration exceeds a few thousand mg Cu/kg (see discussion of plateau response below). Thus, Cu phytotoxicity will seldom be a significant problem during land disposal of dredged materials. But Zn phytotoxicity is a potential problem whenever Zn-rich soils become acidic. Rainfall leaching and N-fertilizers acidify upland soils with time. Further, study using multiple toxic metals makes assessment of causality more difficult. By use of the Zn-rich sludge, phytotoxicity can be expected to result only from added Zn.

As originally reported by Chaney et al. (1982), and recently reviewed by Corey et al. (1987) plant metal concentration approaches a plateau with increasing sludge application rate for a sludge. This happens because the sludge adds not

only metals, but also specific-metal-adsorption capacity. At low sludge application rates, soil specific-metal-binding-sites hold metals more strongly than the sludge metal-adsorption-sites. However, as metal concentration increases, the soil metal-adsorption-sites become saturated in the soil-sludge mixture as sludge application rate increases. Above low sludge application rates, the sludge metal adsorption sites control metals bioavailability in the soil-sludge mixture. Another implication of this model is that plant metal concentration is an increasing curvilinear function of increased sludge metal concentration at equal metal application rates (other factors unchanged). Supporting this model, Zn toxicity was induced in all species at higher levels of soil Zn in the present study using a sludge very high in Zn, but Zn phytotoxicity has not resulted at pH ≥ 5.5 with even high loading rates of median quality sewage sludges (≤1500 mg Zn/kg).

A further goal of the experiment was a comparison of different cultivars of several grasses for use in revegetating acidic soils with excessive Zn. Zinc toxicity limits growth of many species around many smelters and non-ferrous mine waste storage areas in the U.S. and other countries (Beyer, 1988; Chaney et al., 1988; Oyler, 1988). Many dredged materials are very rich in Zn because Zn is used in many industrial processes. For example, the Corpus Christi, TX, and Takoma, WA harbor sediments are very rich in Zn.

Zinc toxicity has been observed in acidic soils as a result of Zn contamination from a variety of sources. A few soils are enriched in Zn and other elements because they developed on an outcropping of a metal deposit, while others were contaminated by seepage from ore bodies (Cannon, 1955; Staker, 1942; Staker and Cummins, 1941), or dispersal of ore or tailings (Takijima and Katsumi, 1973; Kobayashi, 1978). Zn toxicity was observed in cotton and soybean after accumulation of Zn from pesticide sprays used in peach production (Lee and Page, 1967; Lee and Craddock, 1969). Sewage sludge rich in Zn has caused toxicity in many species (Chaney and Giordano, 1977; Logan and Chaney, 1983; Marks et al., 1980; Williams, 1980; Berrow and Burridge, 1981). Mixing ground rubber (contains 0.5-4% Zn) in potting media, or mixing ash from burning rubber materials into soils has caused phytotoxicity (Milbocker, 1974; Patterson, 1971). Accumulation of Zn from galvanized containers or below galvanized fences or electricity transmission towers has caused Zn toxicity, and even caused selection of Zn tolerant ecotypes of several grasses (Antonovics et al., 1971; Bradshaw, 1977; Foy et al., 1978; Baker, 1981; 1987; Jones, 1983).

Accumulation of Zn from emissions of Zn, Pb, or Cu smelters has caused toxicity in many plant species. For example, emissions of the Zn smelters at Palmerton, PA, have killed or prevent regrowth of natural forests (Beyer, 1988; Oyler, 1988), and caused Zn toxicity in garden crops, lawn grasses, deer, and grazing horses (Beyer, 1988; Chaney et al., 1988). Zinc toxicity to grasses is so severe in the Borough of Palmerton that many homeowners have covered their lawns with stones or other mulch materials. These lawn and garden soils contain as high as 10,000 mg total Zn/kg and 100 mg total Cd/kg, while Cu and other elements are not present at excessive levels. Even additions of limestone to raise soil pH to 7 do not correct Zn toxicity enough to allow Kentucky bluegrass to persist on these soils. Angle et al. (1988) found that mycorrhizal infection aided

plant growth and metal tolerance, and even decreased plant Zn and Cd concentrations compared to uninnoculated controls when high plant levels were reached in the control. Baker and Bowers (1988) evaluated growth of lettuce on Palmerton area soils, noting that high soil Zn from long-term smelter pollution was much less phytotoxic or bioavailable than freshly added Zn salts. After adjusting soil pH and adding fertilizers to smelter polluted soils, time is required for metal fixation processes to proceed in the soil (Bruemmer et al., 1986).

Use of metal tolerant ecotypes of grasses to revegetate soils containing high levels of metals has been described by Bradshaw (1977), Smith and Bradshaw (1972), Humphreys and Bradshaw (1977), Johnson et al. (1977) and Baker (1987). In tolerant "ecotypes" of several species have been selected at Zn mine waste sites in several nations, and genes for Zn tolerance exist in natural populations of many forage species (Walley et al., 1974). Metal resistance mechanisms remain elusive (Taylor, 1987), but Van Steveninck et al. (1987) reported that the Zntolerant grass Deschampsia ceapitosa formed specialized vacuoles in root cortex cells which accumulated high levels of Zn and appear to explain Zn tolerance by this ecotype. The possible role of phytochelatins in metal tolerance by plants remains unclear because these compounds chelate many metals, but ecotype are selected with specific tolerance to a single metal (Baker, 1987).

The metal tolerance red fescue ecotype (and commercial cultivar), 'Merlin', has provided vegetative stabilization of soll in the Zn toxic Blue Mountain at Palmerton if some limestone and fertilizer nutrients are provided (Oyler, 1988; Chaney, et al., 1988). Oyler (1988) also reported some success with tall fescue and some warm season grasses when he added limestone to the soil, and surface applied a mixture of sewage sludge and fly ash to these Zn toxic soils. Because Zn-tolerant species, cultivars, or ecotypes may be a practical solution to revegetation of extensive Zn toxic soils in the vicinity of Zn smelters and other Zn-polluted soil, or may be useful on dredged material areas with excessive soil Zn, we included evaluation of the tolerance of Zn by cultivars of 4 grass species along with the study of Cyperus and other species tolerance and accumulation of metals.

Although little tolerance was found in Kentucky bluegrass or perennial ryegrass, several turf-type tall fescue cultivars and 'Merlin' red fescue offer much promise for grasses which can grow on sites with potentially Zn toxic soils.

## **MATERIALS AND METHODS**

Experiment Design For comparison of metal tolerance and uptake by 'Folsom' nutsedge (Cyperus esculentus L.) and other species, methods developed over the last 10 years to study heavy metals in soil-plant systems were used. It is well known that soluble salts of metals are much more plant available than normal metal species in the environment which have reached quasi-equilibrium with soil adsorption and occlusion sites. Other identified errors in methodology for study of sludge-applied metals were discussed by Chaney et al. (1978), Logan and Chaney (1983), Chang et al. (1987) and Corey et al. (1987). For example, fresh sludges are rapidly biodegraded, causing a temporary increase in soluble metals in soil, and higher metal uptake by plants. Also, rapid mineralization and nitrification of applied sludge-N can rapidly lower soil pH.

To avoid these problems, sludge was obtained from a city with substantial Zn-pollution from a hot-dip galvanizing factory. The sludge had been applied to sand beds and allowed to remain on the sand beds subject to rainfall for three years. This allowed much of the sludge N to be mineralized and leached, and for sludge organic matter to be extensively stabilized. The acidic pH of the stabilized sludge was desired to allow higher potential uptake of Zn and a reliable test of relative Zn tolerance.

The experiment was viewed as a factorial interaction of crop species with rate of sludge-Zn application, and used a randomized complete block design with three replications. Each block was placed on one greenhouse bench.

Plant Germplasm One cultivar of most crops were grown under identical conditions for comparison with the response of Cyperus to sewage sludge application rate. Besides Cyperus, the other species included were 'Williams' soybean (Glycine max Merr. L.), 'Klages' barley (Hordeum vulgare L.), 'Paris White Romaine' lettuce (Latuca sativa L., var. longifolia), 'Reubens' Canada bluegrass (Poa compressa L.), and 'Aldous' little bluestem (Schizachyrium scoparium). Certified seed were obtained for each cultivar (see acknowledgment). Six to nine cultivars (see Results) of four grass species were studied for potential Zn resistance: red fescue (Festuca rubra), Kentucky bluegrass (Poa pratensis), tall pascue (Festuca arundinacea), and perennial ryegrass (Lolium perenne).

Yellow nutsedge tubers were obtained from plants maintained to produce tubers. The original tubers were provided by Dr. B.L. Folsom (U.S. Army Engineers, Vicksburg, MS), and are designated cultivar 'Folsom'. After trying several methods to maintain this vegetatively reproduced plant species, plants were grown in a peat-vermiculite potting medium with adequate amounts of required nutrients (Pro-Mix B). During growth, the media were fertilized with a soluble horticultural fertilizer (20-20-20 plus microelements, plus added 1 mM MgSO<sub>4</sub>) once per two weeks to avoid nutrient deficiency. Long day conditions (16 hr.) were maintained to minimize flowering and maximize tuber production. When tubers were needed for an experiment, they were harvested from the potting media, washed with deionized water, and trimmed to remove the rhizome tissue which had connected the tuber to the parent plant. After trimming and washing,

tubers were maintained in aerated 0.5 Hoagland macro nutrient solution (2.5 mM Ca(NO<sub>3</sub>)<sub>2</sub>, 2.5 mM KNO<sub>3</sub>, 1.0 mM MgSO<sub>4</sub>, and 0.1 mM KH<sub>2</sub>PO<sub>4</sub>) for 48 hours before planting to remove putative germination inhibitors. Suspending the tubers in the aerated nutrient solution allowed immediate germination, with one shoot per tuber. Before adopting this approach to maintaining Cyperus tubers, several laboratories which used yellow nutsedge in weed science research were contacted, and several methods they recommended to store harvested tubers were evaluated. Short-term air drying of the tubers to inhibit spoilage and allow refrigerated storage caused most tubers to die. No practical storage method was found for this strain of yellow nutsedge, although some other strains may allow storage. Excessive drying or freezing are known to kill Cyperus (Jansen, 1971; Wills et al., 1980; Day and Russell, 1955; Thomas, 1969; Folsom et al., 1981; Holm et al., 1977).

Soil Properties The soil selected for the study was Sassafras sandy loam. Sassafras is a fine-loamy, mixed, mesic, Typic Hapludult. This soil was selected because it had been used in several studies of metal phytotoxicity and uptake conducted in this laboratory (Chaney et al., 1978; White et al., 1979a, 1979b, 1979c; White and Chaney, 1980), and because it has a relatively low ability to adsorb metals. Further, it was available in a naturally strongly acidic condition such that the soil-sludge mixture could easily be adjusted to pH 5.5. The low pH Sassafras was obtained from an acidic field of Sassafras sl on the Beltsville Agricultural Research Center. Part of the field has been protected from pesticide applications and maintained for use in greenhouse pot research.

Since the original studies using Baltimore sludge (Chaney et al., 1978) or ZnSO<sub>4</sub> (White et al., 1978a; 1979c) and air-dried Sassafras sandy loam, it has become more common to try to use field moist, non-dried soil, to help keep the soil in a chemical and biological condition which reflects the field condition (Ross and Bartlett, 1981). Thus, the fresh soil was collected from the field and it was not air-dried. The field-moist soil was sieved (< 2 mm), mixed well, and stored in closed polyethylene bags to retain the moisture. After sieving, the Sassafras sl was pH 5.3 (1:1, soil:water; 1 hr. incubation), contained 1.2 % organic matter, and had cation exchange capacity of 54 mmol charge/kg soil (5.4 meq/100 g) (White and Chaney, 1980). The initial moisture content (8.8% on a dry soil basis) was obtained to allow calculation of the correct amounts of soil and sludge for preparing the soil-sludge mixtures.

Sludge Properties As noted above, it was also necessary to carefully select the sewage sludge to avoid errors in study of sludge-applied metals. The sludge needed to be acidic enough so that the sludge-soil mixtures would reach the desired pH, near 5.5. The sludge was obtained from Wilmington, OH, where it had been on open sand drying beds for about 3 years. Soluble salts were leached, the organic matter had become well stabilized, and much of the mineralizable N had been removed. Further, the highly stabilized high Zn sludge had a peat-like texture and appearance and was at low pH (5.0). The sludge contained about 43,500 mg Zn/kg dry weight (Table 1). Much of this Zn was known to have come from a hot-dip galvanizing factory. Other elements were present at the low levels of most

Domestic sludges, except Pb which was nearly as high as Zn. The collected air dry sludge was sieved (<2 mm) and mixed well before weighing out portions to mix with soils.

Table 1. Characteristics of high-Zn sludge prepared for use in the experiment.

Sludge	Zn	Cd	Cu	Ni	Fe	рН
	******		<b>m</b> g/kg	DW		
High-Zn	43500	11.2	700	23	3.02	5.02

Treatments. The experiment was designed as a factorial interaction study of the effect of crop species and sludge-Zn addition rate on crop response and composition. Sludge-Zn addition rates used were 0, 2, 4, and 8% sludge addition by dry weight which supplied approximately 0, 870, 1740, and 3480 mg Zn/kg soil-sludge mixture. These levels were selected based on our earlier research (Chaney et al., 1978; White and Chaney, 1980), so that Zn phytotoxicity should be observed in most crops at some sludge rates. The sludge decreased the bulk density of the amended soil so that less weight of

Table 2. Amounts of soil and sludge required for each pot for the several sludge-Zn application rates<sup>1</sup>. Moist soil contained 8.8% water on a dry soil basis [(dry soil weight)-1.088 = moist soil weight].

Sludge Rate	Zinc Rate	Moist Soil	Air-Dry Soil	Sludge	Total
%	mg/kg	g/pot		dry g/pot	*******
0	1.3	. 1958	1800	0	1800
2	870.	1834	1686	34.4	1720
- 4	1740.	1692	1555	64.8	1620
8	3480.	1531	1408	122.4	1530
	Rate % 0 2 4	Rate Rate  % mg/kg  0 1.3 2 870. 4 1740.	Rate Rate Soil  mg/kg g/pot  1.3 1958 2 870. 1834 4 1740. 1692	Rate Rate Soil Soil  2 mg/kg g/pot  0 1.3 1958 1800 2 870. 1834 1686 4 1740. 1692 1555	Rate Rate Soil Soil  2 mg/kg g/pot dry g/pot  0 1.3 1958 1800 0 2 870. 1834 1686 34.4 4 1740. 1692 1555 64.8

Fertilizer salts were added to assure appropriate fertility for pot experiments with Sassafras sl. Application rates and sources of added fertilizer nutrients were; 100 mg N/kg dry soil as  $Ca(NO_3)_2 \cdot H_2O$ ; 400 mg P/kg as  $KH_2PO_4$  plus  $CaHPO_4$ ; 252 mg K/kg as  $KH_2PO_4$ ; 200 mg Mg/kg as  $MgSO_4 \cdot 7H_2O$ ; 2.62 mg Zn/kg as  $ZnSO_4 \cdot 7H_2O$ ; 1.28 mg Cu/kg as  $CuSO_4 \cdot 5H_2O$ ; 1.00 mg B/kg as  $H_3BO_3$ ; and 0.18 mg Mo/kg as  $Na2MoO_4 \cdot 2H_2O$ .

amended soil provided the same volume as 1.8 kg of dry Sassafras sandy loam (Table 2). Equal volumes of soil or soil-sludge mixture were placed in all pots.

Aliquots of solutions of the fertilizer salts were added to the moist sieved soil in pots. After mixing fertilizers and high Zn sludge with the moist Sassafras sandy

loam soil, the mixtures were returned to 15 cm polyethylene pots (with drainage holes covered with 44 mesh polyethylene screen). The mixed soils were watered to field capacity and incubated 7 days. This allowed the mixtures to react with the soil and reach a quasi-equilibrium before remixing the moist soil-sludge mixtures. After the second mixing, tubers or seeds were planted.

Plant culture Appropriate numbers of tubers or seeds of each crop species were sown in the pots, and large seeded species were thinned to uniform numbers of seedlings per pot. The numbers of tubers or seeds were: seven pregerminated yellow nutsedge tubers; 10 seeds of soybean or lettuce; 25 seeds of barley; 25 seeds of little bluestem; and 300 to 500 seeds from each of the 32 grass cultivars noted above (Kentucky bluegrass, 1/4 teaspoon = 300 to 400 seeds; other species, 1/2 teaspoon = 400 to 500 seeds) and of Canada bluegrass. To reduce crusting of the soil surface, the pots were watered so that the seeds were wetted primarily by capillary action, and the pots were covered with paper for 5 to 7 days until seedling emergence to inhibit drying of the soil surface. Average daily temperature in the greenhouse during the experiment ranged between 20 and 28° C, while the daily humidity ranged between 52 and 90%. Seeds in block 2 germinated less well than in blocks 1 and 3, and yields of the small seeded grasses were usually smaller in block 2. Using the block design for the experiment removes this source of variance in the evaluation of results.

Direct seeded lettuce, soybean, and little bluestem germinated poorly and quickly died in the high-Zn treatments. To promote a fairer comparison of relative crop response, these crops were germinated in standard seed germination papers and transplanted to the pots for all sludge rates. Lettuce was thinned to 5 plants per pot, and soybean to three.

The development and symptoms of Zn phytotoxicity in the plants were observed during plant growth. Numerical scores for stunting of the plants, and of chlorosis severity, were assigned to each pot on several dates. Each pot of a perennial crop was clipped twice during the growth. Tall fescue and perennial ryegrass were grown for 34 days after planting for the 1st harvest, and 36 days regrowth for the 2nd harvest. Red fescue, Kentucky bluegrass, and Canada bluegrass were grown for 60 days after planting for the 1st harvest, and 41 days regrowth for the 2nd harvest. The grass plants were clipped at 2 cm above the soil surface at each harvest. Annual crops were grown once starting during the first growth period of the perennial crops, as was little bluestem due to slow growth of this species.

After the first clipping, each pot of the perennial crops received 200 mg N as NH<sub>4</sub>NO<sub>3</sub>. This form of N was used to minimize the change in soil pH due to plant growth and uptake of NO<sub>3</sub>-N.

<u>Plant analyses</u> The clippings were dried at 70° in a forced draft oven, and weighed to measure yield per pot. The dried grass plants were ashed in 200 ml Pyrex beakers at 480° C for 14 hours and the ash dissolved in 3 ml of conc. HNO3 and heated to near dryness on a hotplate. The whole sample was ashed except for barley and Cyperus samples which yielded over 10 grams dry matter per pot; in

these cases, the samples were ground in a stainless steel Wiley mill, and 5 g portions ashed. Subsequently, 10 ml of 3N HCl was added to dissolve the ash, refluxed for 2 hr., and filtered. Zn, Fe, and other elements were analyzed by atomic absorption spectrophotometry. For Cd, Ni, and Pb, deuterium background correction was used.

Soil analyses The analysis of soil pH was obtained for each pot at the end of the growth period (two clippings for the perennial crops). The pH of a 1:1 soil:deionized water slurry was measured after 1 hr. incubation with periodic stirring. Soil samples were obtained from three cultivar entries at the termination of the experiment for analysis of DTPA-extractable metals (Lindsay and Norvell, 1978). Further, water extractable metals were determined after 2 hr extraction with 15 g dry soil/30 mL deionized water.

Statistical Analysis Statistical analysis was conducted using the SAS statistical software. The GLM procedure was used for analysis of variance (factorial model: crop, rate, crop•rate, and block), and means were separated using the Waller-Duncan K-ratio t test. Several variables required logarithmic transformation before analysis to maintain homogeneity of variance across treatments, and the results are presented as geometric means in tables. Linear regression was used to evaluate results of rates of application. A number of outliers in shoot Fe concentration were deleted because they were believed to have been contaminated with soil during harvest. Samples containing over 300 mg Fe/kg were deleted if replicates were much less than this value.

The use of different crops with different yield potentials made evaluation of simple yield results of little value. In order to provide a more useful comparison among crops of the effect of treatments on crop yield, a normalized yield was calculated: the yield results for 2, 4, and 8% treatments for a cultivar were divided by the arithmetic mean yield for the 0 % sludge (control) treatment. This allows the natural variance of yield to be retained for each cultivar or species, but expresses the yield results as % of control, allowing valid comparison across cultivars and species tested.

# **RESULTS AND DISCUSSION**

Soil Analysis Soils were extracted with the normal DTPA solution of Lindsay and Norvell (1978). This procedure is used by many researchers because it is usually well related with metal uptake by plants. However, this method is insensitive to soil pH, the most important variable which affects Zn and Cd uptake by plants.

At the end of the growth period individual pots differed somewhat in soil pH, and this variation was reflected in Zn extracted with deionized water (Table 3). However, the variation among pots in DTPA extractable Zn was small. DTPA used at the standard 1 g soil/2 mL solution is capable of extracting about 654 mg Zn/kg (10 mmol/kg) assuming the 1 Zn:1 DTPA chelate is formed. The standard DTPA method was not able to extract the normal fraction of total Zn at the 4 and 8% sludge rates. Somewhat more moles of Fe + Zn + Mn were extracted than 10 mmol/kg at the 4% and 8% sludge rates. DTPA can form a chelate with 2 metal ions per mole, but only a fraction of the DTPA forms Zn<sub>2</sub>DTPA under these conditions. At these very high levels of total soil Zn, the DTPA became saturated with Zn. Because of this, DTPA extractable Fe declined at the 8% sludge rate because Zn filled the DTPA and prevented Fe extraction. DTPA extractable Mn varied somewhat among sludge rates, and water extractable Mn increased with sludge rate.

Table 3. Zinc, Fe and Mn extracted by DTPA or deionized water, and total metal concentrations from cropped soils after the final harvest. Values are the mean of 6 replications (all pots from cultivars 1 and 2).

Sludge		Zn			Fe		Mn		
rate	DTPA	Water	Total	DTPA	Water	Total	DTPA		Total
<del>-</del> %			mg	extrac	ted/kg	dry soil			
0	4.8	1.0	98	52.4	4.60	3520	3.90	0.10	83.4
2	420.	13.2	916	58.3	0.40	3820	6.50	1.20	96.1
4	655.	34.7	1790	52.7	0.60	4060	8.50	2.10	97.1
8	788.	82.5	3810	11.3	0.30	4550	5.20	3.70	98.6

Soil pH results are shown in Table 4. By harvest or second harvest, soil pH had usually risen in treatments (cultivar and sludge rate) which allowed plants to grow well. The plants were initially supplied NO<sub>3</sub>-N, and plant uptake of this chemical form of N causes rise in soil pH. There was little change in soil pH at the higher sludge rates for soybean and lettuce compared to the initial pH of the soil-sludge mixtures, about 5.3.

<u>Plant Response</u> The relative tolerance and uptake of soil heavy metals varied widely among the different crop species. All crops were grown during the first growth period, while perennial grasses were clipped a second time after regrowth.

The comparison of crop species at each clipping is discussed here, while evaluation of first vs. second clipping is reported below. Where 6-8 cultivars were grown for a crop species, the mean of the response of cultivars in this part of the report, except for 'Merlin' red fescue which behaved so differently from other red fescue lines that results for 'Merlin' are reported separately in the crop comparisons. Discussion of cultivar differences (other than 'Merlin') is reported below. It is important to consider that because of the wide range of yield among entries, the variance was unequal for cultivars and logarithmic transformation was used to obtain valid comparisons of yield and shoot Zn results. Geometric means are shown in the tables, and the Waller-Duncan mean separation letters are for the logarithmic values.

Table 4. Final soil pH of pots cropped with various crop species, and amended with four rates of high-Zn sludge.

		Final	soil pH a	fter one o	r two clips	ings
Стор	N	All Rates	0%-sludge	2%-sludge	4%-sludge	8%-sludge
Red Fescue	81	5.49 fg <sup>1</sup>	5.74 d-q <sup>2</sup>	5.51 klm <sup>2</sup>	5.41 m-p	<sup>2</sup> 5.31 pg <sup>2</sup>
Merlin	12	5.64 bcd	5.83 bcd	5.60 h-1	5.57 h-1	5.54 1-1
K. bluegrass	90	5.58 de	5.84 bcd	5.58 h-1	5.51 k-n	5.38 nop
Tall Fescue	96	5.63 cd	5.78 c-f	5.67 f-i	5.60 h-1	5.48 1-o
P. ryegrass	- 84	5.70 b	5.91 ab	5.76 c-f	5.62 g-k	5.50 k-o
Canada Bluegrass	12	5.52 ef	5.84 bcd	5.49 1-o	5.55 i-1	5.30 pq
Little Bluestem	12	5.57 de	5.81 b-e	5.62 g-k	5.50 k-o	5.37 op
Soybean	12	5.45 g	5.69 e-h	5.51 k-n	5.37 op	5.22 gr
Romaine Lettuce	9	5.35 h	5.55 i-1	5.38 op	5.33 pq	5.15 r
Yellow Nutsedge	12	5.79 a	6.01 a	5.83 bcd	5.77 c-f	5.54 jkl
Barley	12	5.69 bc	5.87 bc	5.74 d-g	5.66 f-j	5.48 1-o
All Crops	110		5.81 A <sup>3</sup>	5.62 B	5.54 C	5.41 D

<sup>&</sup>lt;sup>1</sup>Means in this column followed by the same letter are not significantly different (P  $\leq$  0.05) according to the Weller Duncan K-ratio t test using a factorial model. <sup>2</sup>Means in these four columns followed by the same letter are not significantly different (P  $\leq$  0.05) according to the Waller-Duncan K-ratio t test using the interaction term crop\*rate.

Results of yield, symptom, and leaf composition for crop species (averaged over cultivars and four high Zn sludge rates) are shown in Tables 5 and 6. Striking differences were observed among crop species as the soil Zn level increased. Table 6 shows the means for the 2, 4, and 8% sludge rates so that the non-phytotoxic control treatment results do not dilute the effects of sludge-Zn on crop

<sup>&</sup>lt;sup>3</sup>Overall sludge rate means followed by the same upper case letter are not significantly different ( $P \le 0.05$ ) according to the Waller-Duncan K-ratio t test using a factorial model.

comparisons. The immediately following discussion is based on results in both tables unless specific mention of a Table number is made.

'Merlin' red fescue tolerated the high soil Zn level best of all entries. Romaine lettuce was the most Zn-sensitive crop species, with 99% yield reduction averaged over sludge rates (geometric mean normalized yield). Soybean was next most sensitive, followed by Canada bluegrass, red fescue, Kentucky bluegrass, little bluestem, Perennial ryegrass, yellow nutsedge, barley, and tall fescue. Tall fescue and barley were more tolerant that the other crops excepting 'Merlin' red fescue.

Photographs of representative cultivars are shown for each crop species. For species where 6-8 cultivars were studied, only relatively tolerant and

Table 5. Effect of crop species on yield, symptoms, and composition of the first clipping leaves of 11 crops grown on 4 rates of high Zn sludge-amended Sassafras sandy loam soil at strongly acidic pH.

		Geo. Mean	Geo. Mean	•	Chlorosis	Terminal	Ge	ometric Me	an
CROP	N	Yield	NormYield	Height	severity	Soil pH		Shoot Mn	
		g/pot	%-Control	cm	Green=1		mg/	kg dry wei	ght
First Clipping					•				
Red Fescue	81	0.685 f	26.1 ef	14 e	1.6 cde	5.49 fg	814 d	65.8 b	77.0 a
Merlin red fescue	12	3.91 b	86.8 a	16 e	1.2 e	5.64 bcd	156 h	43.6 d	53.5 de
Kentucky Bluegrass	90	1.34 e	37.4 de	10 f	1.5 de	5.58 de	555 fg	34.1 e	72.2 ab
Tall Fescue	96	2.25 c	75.2 ab	26 c	1.5 de	5.63 cd	830 cd	54.4 c	60.2 b-e
Perennial Ryegrass	84	2.17 cd	51.5 bcd	24 d	1.9 cd	5.70 b	826 cd	53.4 c	70.9 abc
Canada Bluegrass	12	2.16 cd	25.7 ef	10 f	2.0 c	5.52 ef	730 de	53.5 c	49.9 ef
Little Bluestem	12	1.40 de	49.8 cd	•	•	5.57 de	6 <u>44</u> ef	129. a	41.6 f
Soybean	12	0.312 g	19.6 f	•	3.4 a	5.45 g	949 c	150. a	55.7 de
Romaine Lettuce	9	0.029 h	3.7 g	•	3.7 a	5.35 h	2470 a	150. a	78.7 a
Cyperus	12	7.48 a	44.1 cd	64 a	. 2.5 b	5.79 a	499 g	54.5 c	59.1 cde
Barley	12	4.43 5	65.9 abc	58 b	1.3 e	5.69 bc	1290 Ď	32.8 e	61.7 bcd
Second Clipping					. •	·		·	
Red Fescue	67	0.48 d	54.4 cd	42 a	2.4 d		527 b	111. a	
'Merlin'	11	1.41 bc	151. a	29 b	1.1 f	•	170 e	76.9 bc	
K. bluegrass	92	1.72 b	50.2 cd	26 bc	1.9 e		433 cd	69.4 bc	•
Tall Fescue	95	3.69 a	62.7 bc	25 c	2.6 c		516 b	60.7 c	
P. ryegrass	83	1.70 b	39.7 d	21 d	3.5 a		645 a	72.5 bc	
C. bluegrass	10	0.98 c	43.7 d	20 d	3.2 b		472 bc	80.5 b	
Cyperus	12	1.80 b	74.0 b	•	. •	•	376 d	75.1 bc	

Means followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan mean separation.

Table 6. Difference among crops in tolerance of high soil Zn concentrations. Means of yield, chlorosis, and shoot composition for sludge treatments only (2, 4, and 8% additions).

<del></del>		Geome	tric	Geo.	Norm.	Geomet	ric	Shoo	ot	Chloros	is	Heigh	nt
CROP	N		Yield			Shoot		Mn		Score			
		g/p	ot	%-Con	trol		-mg/k	cg DW-		Green-	-1	cm	
First Clipping	1:												
Red Fescue	61	0.46	e	17.2	ef	965	cd.	71.0	d	1.8 €	ef	12.8	е
Merlin	9	3.73	ab	83.0	a .	183	g.	39.5	fg	1.2	1	15.8	d
K. Bluegrass	68	1.06	i <b>d</b> .	29.6	de	645		35.3	g	1.6 1	fgh	8.7	f
Tall Fescue	72	2.09	bc	69.8	ab	1060	cd	55.5		1.7 1	fg	25.5	Ь
P. ryegrass	63	1.80	cd	42.9	bcd	1040	cd	58.3			le	22.5	C
C. bluegrass	9	1.38	cd	16.4	f	898	cd	53.4	d-g			8.0	f
Little blueste	<b>m</b> 9	1.11	cd	39.5	bcd	804		153.	c T	•			•
Soybean	9	0.18	f	11.4	f	1120		196.	Ь	4.2 1	)		-
Lettuce	9	0.01		1.2		3620		244.	a	4.6			•
Cyperus	٠ 9		•	33.6		580		60.4	de	3.1		57.3	a
Barley	9			57.6		1660		40.8				57.8	
Second Clippin	ng:												
Red Fescue	60	$\overline{0}.33$	d	21.4	C	1180	ab	129.	a	2.8	d	18.4	C
Merlin	8	1.55	b	110.	<b>a</b> .	339	е	76.1	bc	1.1	f	25.7	Ь
K. Bluegrass	70	1.24	Ь	24.5	С	960	Ċ	76.1	bc	2.2	e	25.9	Ь
Tall fescue	.72	3.08	a	46.6	b	1100	bc	63.3		3.2		40.5	
Per. ryegrass	63	1.10		16.5	cd	1340	a	77.0		4.3		23.5	_
C. Bluegrass	8	0.65		12.1		1140	-			3.9		18.3	
Cyperus	9	1.45		42.0		674		67.7		•	-	•	•

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio t test.

- Figure 1. Photographs of representative cultivars of crops studied. A) 'Folsom' yellow nutsedge (Cyperus); B) 'Williams' soybean; C) 'Merlin' red fescue; D) \_\_\_\_ red fescue; E) 'Houndog' tall fescue (resistant); F) 'Kentucky-31' tall fescue (susceptible); G) \_\_\_ Kentucky bluegrass (resistant); H) \_\_\_ Kentucky bluegrass (susceptible); I) \_\_\_ perennial ryegrass (resistant); J) \_\_\_ perennial ryegrass (susceptible); K) 'Klages' barley; L) 'Parris Island Romaine' lettuce; M) 'Aldous' little bluestem; and N) 'Reubens' Canada bluegrass.
  - Figure 1-A. Effect of increasing rates of high-Zn sludge-applied Zn on growth and chlorosis of 'Folsom' Cyperus. Upper photo from first clipping; lower photo from second clipping.
  - Figure 1-B. Effect of increasing rates of high-Zn sludge-applied Zn on growth and chlorosis of lettuce and soybean. Upper photo, 'Paris White Romaine' Lettuce; lower photo, 'Williams' soybean.
  - Figure 1-C. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of barley and little bluestem. Upper photo, 'Klages' Barley; lower photo, 'Aldous' little bluestem.
  - Figure 1-D. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Reubens' Canada bluegrass. Upper photo at first clipping; lower photo at second clipping.
  - Figure 1-E. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Kentucky-31' tall fescue. Upper photo at first clipping; lower photo at second clipping.
  - Figure 1-F. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Houndog' tall fescue. Upper photo at first clipping; lower photo at second clipping.
  - Figure 1-G. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Manhattan' perennial ryegrass. Upper photo at first clipping; lower photo at second clipping.
  - Figure 1-H. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Merion' Kentucky bluegrass. Upper photo at first clipping; lower photo at second clipping.
  - Figure 1-I. Effect of increasing rates of sludge-applied Zn on growth and chlorosis of 'Merlin' red fescue. Upper photo at first clipping; lower photo at second clipping.

relatively susceptible cultivars are shown (Figure 1). As sludge rate increased, all crops suffered some stunting or chlorosis except 'Merlin' red fescue. At the two higher sludge Zn rates, severe chlorosis was seen in Cyperus, in perennial ryegrass cultivars, in Kentucky and Canada bluegrass, and in soybean and lettuce.

At higher soil Zn levels, some crops with high foliar Zn concentration suffered severe chlorosis, and had large yield reductions, indicating severe Zn phytotoxicity. Yellow nutsedge and Canada bluegrass had the most severe chlorosis at 4 and 8% sludge among the monocots (Table 5; Figure 1), and among the dicots, lett. ce and soybean chlorosis were most severe (Table 5; Figure 1). Perennial ryegrass and Kentucky Bluegrass also had severe chlorosis at higher sludge rates, while tall fescue had lower chlorosis severity and 'Merlin' red fescue remained green at all treatments. Thus, there was a reasonable correlation between chlorosis severity and yield reduction.

The Zn concentration in the leaves varied widely among crops at each sludge-Zn rate. Romaine lettuce accumulated the highest Zn concentrations (geometric mean across sludge Zn levels), followed by barley, soybean, tall fescue, perennial ryegrass, red fescue, Canada bluegrass, little bluestem, Kentucky bluegrass, and yellow nutsedge, with 'Merlin' red fescue containing the lowest Zn level. The ability of 'Merlin' to exclude Zn is remarkable compared to other red fescue cultivars, and to other crop species. During regrowth of perennial species, the shoot Zn was higher than during the first growth period, as were chlorosis severity and yield reduction.

In comparing the apparent tolerance of Zn by crop species, one must consider at least two factors: 1) uptake of Zn by the crop (exclusion of Zn allowing resistance to soil Zn), and 2) degree of yield reduction or toxicity severity at a particular level of foliar Zn (physiological Zn tolerance) (Baker, 1987; Foy et al., 1978). For example, White et al. (1979a) found that different soybean cultivars differed in both relative Zn uptake to leaves, and extent of yield reduction at 500 mg Zn/kg leaves. In contrast with excessive soil Cu which appears to cause phytotoxicity by effects on root metabolism, excessive soil Zn can cause phytotoxicity by reactions in the plant shoots. White et al. (1979a; 1979b) found that chlorosis induced by excessive soil Zn in strongly acidic soils results from Zn-by-Fe interactions in the leaves of soybean rather than by inhibition of Fe uptake or translocation by the roots. Shoot genotype controlled Zn tolerance, while root genotype controlled Zn concentration in the shoots (White et al., 1979b). In the present experiment, only Canada bluegrass had such low shoot Fe that its chlorosis might be presumed to be related to shoot Fe concentration, per se.

Information on Zn tolerance and uptake by many crop species was reported by Boawn and Rasmussen (1971) and Boawn (1971). Their studies used neutral or calcareous soils, with addition of ZnSO<sub>4</sub> to air-dried soils. Fortunately, the pH buffering of these soils was high, so little pH reduction occurred due to the Zn addition (saturated paste pH fell from 7.5 to 7.0). Under the pH conditions of that study, *Poaceae* species were more sensitive than many dicotyledonous species, including legumes and vegetables (although no chlorosis was observed in any species in their study) (Table 7). Significant yield reduction began at 400 to 700 mg Zn/kg leaves, while the concentration generally agreed to clearly indicate Zn-

phytotoxicity is about 500 mg/kg (Chaney and Giordano, 1976; Loyan and Chaney, 1983; Sommers, 1980).

It is believed that the ability of Zn and other heavy metals to be phytotoxic to *Poaceae* species at high soil pH is due to their use of the phytosiderophore mechanism to absorb soil Fe. At higher soil pH, heavy metals (Cu, Zn) are able to displace Fe from Fe-mugineic acid (Mino et al., 1983), or keep mugineic acid from chelating soil Fe (Takagi et al., 1988; Treeby et al., 1989). Thus, these species would be relatively more susceptible to soil Zn at high pH compared to other plant families (those not using the phytosiderophore-mechanism to obtain soil Fe), than at acidic pH. In the present study, the *Poaceae* species were clearly more tolerant of Zn in acidic soils than were the non-*Poaceae* species, the pattern commonly observed in Zn toxic fields and gardens.

Table 7. Comparison of Zn uptake and tolerance by different crop species on neutral pH soil amended with 0-500 mg Zn/kg as ZnSO<sub>4</sub> (Boawn and Rasmussen, 1971).

Crop	Zn mg Zn/kg soil=	in sho	oots, mg/k 500	g Yield Decrease at P < 0.05	- <del></del>
		mg/k	cg DW	*	<del></del>
Monocots				•	
Field Corn		484	763	26	
Sweet Corn		475	713	29	
Sorghum-1		748	1030	15	
Sorghum-2	•	646	1140	19	
Barley		910	2110	15	
Wheat		522	909	10	
Dicots					
Field Bean		151	257	-	'
Snapbean		111	213	-	
Alfalfa		142	345	13	
Clover		161	252	•	
Pea-1	•	285	489		
Pea-2		236	522	2?	٠.
Lettuce		250	665	27	
Spinach	•	640	945	10	
Potato-1		163	327	-	
Potato-2		138	346	••• · · · · · · · · · · · · · · · · · ·	
Sugarbeet		509	1070	22	
Tomato		316	514	24	

Other authors have reported Zn phytotoxicity at somewhat lower foliar Zn concentration than found by Boawn and Rasmussen (1971). Davis and Beckett (1978) evaluated metal phytotoxicity in barley using sand culture, and reported Zn phytotoxicity at as little as 300 mg Zn/kg shoots. Sand culture may not be an appropriate method to assess metal phytotoxicity because the metals are at such high concentration in the interstitial water after evapo-transpiration by the plants. Soil studies generally support the findings of Boawn and Rasmussen on the concentration of Zn in leaves when phytotoxicity begins.

After evaluating the crop comparisons of Table 5, it was apparent that some crops (e.g., lettuce) had too high normalized yield because the control crop was included in the "crop" averages. Thus, it was considered appropriate to recalculate the "crop" means and statistics using only the high Zn-sludge treatments (2, 4, and 8% added sludge) (Table 6). In this comparison, the low Zn concentration in the controls, the low chlorosis scores of the control, and high normalized yield of the controls does not confound interpretation of the "crop" differences in Zn-tolerance and Zn uptake to shoots.

In Table 6, some crops are clearly more susceptible to soil Zn than others (lettuce and soybean are very susceptible, while barley, tall fescue, and Merlin red fescue are relatively resistant). 'Merlin' red fescue resisted soil Zn better than the other entries. Tall fescue appears to offer promise for culture on high Zn soils, especially several cultivars (see below). Perennial ryegrass, Kentucky bluegrass, Canada bluegrass, and little bluestem had little resistance to excessive soil Zn, while the crop plants soybean and lettuce were very intolerant.

Also, the crops differed in relative uptake of Zn to the shoots (lettuce highest, barley ... K. bluegrass, Cyperus, and 'Merlin' red fescue), and in relative yield reduction at a particular shoot Zn concentration. In this experiment, the relation of Zn concentration in leaves and yield reduction was close for most crops, but a few crops appeared to differ from this pattern. The relationship of yield reduction and Zn concentration is shown in Figure 2. This Figure shows the geometric mean normalized yield (for the 2, 4, and 8% sludge rates) plotted against the geometric mean shoot Zn concentration. By using the normalized yield for only the sludge-Zn treatments, the relative yield reduction for each cultivar is used in the calculation, normalizing the estimate across crop entries. By using the geometric means, the effect of any single cultivar or sludge rate has less effect on the result.

In Figure 2, the crops Merlin red fescue, yellow nutsedge, Kentucky bluegrass, red fescue, and Canada bluegrass lie on a line (the line shown is from linear regression of the logarithm of shoot Zn vs. the logarithm of normalized shoot yield for those crops). Lettuce was so severely injured by Zn that its position can not be interpreted. Tall fescue and barley lie far to the right of and above the line for the other crops, while perennial ryegrass and little bluestem lie somewhat to the right of the other crops.

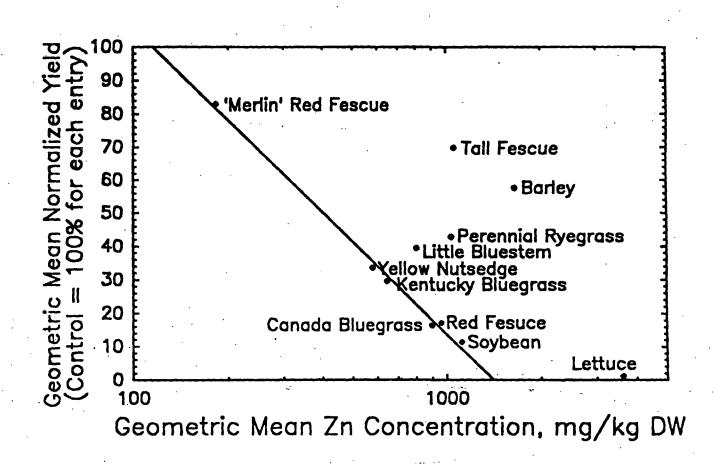


Figure 2. Relationships of geometric mean shoot Zn concentration and geometric mean normalized yield for 11 crop species (averaged across 2%, 4%, and 8% sludge Zn rates, and over 1-8 cultivars per crop.

This indicates that barley tolerated much more leaf Zn than did other crop species. Further, the unusual tolerance of soil Zn by 'Merlin' red fescue is clearly shown to result from the ability of this cultivar (ecotype) to exclude Zn compared to other red fescue cultivars and other crop species. A similar conclusion was drawn from Zn concentration in 'Merlin' red fescue growing on extremely Zn polluted soils on Blue Mountain at Palmerton, PA (Chaney et al., 1988). 'Merlin' excluded Zn and Cd compared to most other crop species and even excluded Cd relative to Zn. This makes 'Merlin' better at protecting wildlife from excessive soil Zn than other crops which appear to resist excessive soil Zn (e.g., tall fescue).

Crop species interaction with sludge-Zn rate Tables 8 and 9 report the full treatment rate and crop species means for analyzed data from the first clipping and Tables 10 and 11 report the data for the second clipping. These Tables allow cross crop comparisons at the different sludge rates, and make possible the identification of crops which are more tolerant to soil Zn. The geometric mean normalized yield allows judgement of metal tolerance (Tables 8 and 10) while Tables 9 and 11 allow comparison of Zn uptake relationships. Rather than the crop means across sludge rates discussed above, the whole data of crop and sludge-Zn rate allow discussion of responses. Here one can readily see that 'Merlin' is quite resistant to soil Zn, Cyperus intermediate, and lettuce and soybean quite sensitive.

Second Clipping shoots of perennial crops After harvesting the initial growth of shoots of Cyperus, red fescue, Kentucky bluegrass, tall fescue, perennial ryegrass, and Canada bluegrass, the soils were fertilized with NH<sub>4</sub>NO<sub>3</sub> (200 mg N/kg soil) to support regrowth of these crops. This was desired because field experience in Palmerton, PA (Oyler, 1988; Chaney et al., 1988) and the greenhouse work of Walley et al. (1974) showed that often seed sown in highly Zn toxic soils germinated and grew for a while, but only metal tolerant cultivars or ecotypes persisted with repeated harvest or clipping.

Comparison of First and Second Clippings of Perennial Crops Comparison of Tables 8 and 9 with 10 and 11 indicated that a specific comparison of normalized yield, Zn uptake, and chlorosis score for the first and second clippings would be useful. Several species had more severe chlorosis in the second clipping growth period, and higher shoot Zn than in the first growth period. Thus, Table 12 includes these specific comparisons in the crop-by-rate format to allow full comparison of changes between the clippings. Indeed, chlorosis in more severe in nearly each crop, shoot Zn in increased in most cases, and normalized yield is reduced for most of the crops, but not 'Merlin'. Genetic metal tolerance is maintained during growth and regrowth, while normal red fescue and perennial ryegrass cultivars became less hardy with time and would not have been expected to resist environmental pressures in a natural Zn-rich soil habitat.

Table 8. Effect of crop species interaction with rate of high Zn sludge on yield, normalized yield (% of Control within crop), phytotoxicity symptoms, and composition of the first clipping leaves of 11 crops.

Crop	Rate N		Geometric Mean NormYield	Chlorosis	Shoot Height	
		g/pot	%-Control	1=Green	CIII	
Red Fescu	0 20 2 20 4 21 8 20	0.906 j-n 0.518 nop	92.8 ab 33.7 e-j 20.0 h-k 7.5 lmn	1.0 k 1.2 jk 1.6 gh 2.7 d	18.1 jk 14.0 l-o 12.8 m-p 11.5 n-r	
Merlin red fescu	0 3	4.477 c-f	99.5 a 67.9 a-e 100.6 a 83.8 a-d	1.0 k 1.2 ijk 1.2 ijk 1.3 h-k	16.2 klm 15.7 klm	•
Kentucky Bluegras:	0 22 5 2 22 4 23 8 23	2.78 d-i 1.42 h-m 1.41 h-m	77.1 ab 40.6 abc 39.1 a-e 16.6 a-f	1.0 k 1.2 ijk 1.5 ghi 2.2 e	12.4 m-q 9.8 p-s 9.1 qrs 7.3 st	
Tall Fescue	0 24 2 24 4 24 8 24	2.71 d-i 2.11 e-j	94.0 a-d 90.5 c-i 70.5 d-i 53.3 jkl	1.0 k 1.4 g-j 1.7 fgh 2.0 e	27.9 g	
Perennial Ryegrass	0 21 2 21 4 21 8 21	2.84 d-i 1.81 f-k	89.5 abc 67.6 a-e 43.0 b-h 27.1 f-i	1.0 k 1.7 fg 2.1 e 2.9 d	26.9 g 24.7 gh 22.3 hi 20.6 ij	
Canada Bluegras	0 3 s 2 3 4 3 8 3		98.4 a 46.0 a-g 27.6 f-k 3.5 no	1.0 k 1.7 fgh 2.0 ef 3.3 c	15.0 k-n 10.7 o-s 8.5 rst 4.8 t	
Little Bluestem	0 3 2 3 4 3 8 3	1.58 g-l 1.38 h-m	99.8 a 56.0 a-f 48.8 a-g 22.6 g-k	•	•	
Soybean .	0 3 2 3 4 3 8 3	0.256 o-r 0.206 pqr	99.8 a 16.1 jkl 13.0 klm 7.1 mn	1.0 k 3.3 c 4.2 b 5.0 a		
Romaine Lettuce	0 3 2 3 4 3 8 0	0.768 k-n 0.013 st 0.015 s 0.005 t	97.7 a 1.6 o 1.9 o 0.6 p	1.0 k 4.0 b 4.8 a 5.0 a	• • •	
Yellow Nutsedge	0 3 2 3 4 3 8 3	6.94 a 5.96 ab 3.76 c-g 3.08 d-h	99.9 a 94.1 ab 22.2 g-k 18.2 ijk	1.0 k 1.2 ijk 4.0 b 4.0 b	84.7 a 75.7 b 52.3 e 44.0 f	
Barley	0 3 2 3 4 3 8 3	4.12 c-f	98.7 a 42.8 b-h 61.3 a-f 72.7 a-e	1.0 k 1.0 k 1.5 ghi 1.7 fgh	58.7 cd 59.3 c 55.0 de 59.0 c	

Table 9. Effect of crop species interaction with rate of high Zn sludge on normalized yield (% of control within crop), and Zn, Mn, and Fe concentration in first clipping leaves of 11 crops.

rop	Rate N	Geometric Mean NormYield	Geo. Mean Shoot Zn	Shoot Mn	Shoot Fe
		%-Control		g/kg dry wei	ght
ed Fescue	0 20	92.8 ab	46 vw	69 i-m	70 b-i
	2 2	33.7 e-j	662 m-o	57 j-r	78 a-g
	4 2	l 20.0 h-k	942 h-1	84 <b>ij</b>	90 a-e
-	8 2	7.5 lmn	1440 cd	71 1-1	103 a
lerlin	0	3 99.5 <sub>.</sub> a	30 xy	68 i-m	68 c-i
ed fescue	2	67.9 a-e	88 t	41 m-s	51 f-k
	2 4 8	3 100.6 a	165 s	33 qrs	49 f-k
	8 .	3 83.8 a-d	318 qr	43 1-s	52 f-k
(entucky	0 2	2 77.1 ab	42 VW	43 1-s	65 c-i
Bluegrass	2 2		415 pq	20 s	. 75 a-h
•	4 2	3 39.1 a-e	621 m-o	38 o-s	80 a-f
	8 2	3 16.6 a-f	1020 f-j	47 k-s	93 a-d
[a]]	0 2	4 94.0 a-d	49 uv	59 j-q	64 c-i
Fescue	2 2		821 i-m	49 k-r	56 f-1
	4. 2		1070 e-i	67 i-n	62 e-j
	8 2		1370 c-f	51 k-r	62 e-j
Perennial	0 2	1 89.5 abc	48 uv	49 k-r	78 a-g
Ryegrass	2 2		768 j-n	46 k-s	70 b-i
	4 2		1010 g-j	68 i-m	71 a-i
	8 2		1425 cde	61 <b>j</b> -p	71 a-i
anada	0	3 98.4 a	42 VW	66 j-o	43 ijk
Bluegrass	2	3 46.0 a-g	573 no	39 n-s	44 h-k
, 🗸	4	3 98.4 a 3 46.0 a-g 3 27.6 f-k 3 3.5 no	915 h-1	49 k-s	57 f-k
•	8	3 3.5 no	1380 cde	73 ijk	65 c-i
ittle	0	3 99.8 a	26 y	83 ij	56 f-k
Bluestem	2	3 99.8 a 3 56.0 a-f 3 48.8 a-g 3 22.6 g-k	544 op	118 gh	41 ijk
•	4 .	3 48.8 a-g	785 j-m	142 fg	42 ijk
	8	3 22.6 g-k	1220 d-h	200 cd .	32 jk
oybean	0	3 99.8 a	65 u	95 hi	95 abc
	-		713 1-0	163 ef .	52 f-k
	4	3 13.0 klm	1110 d-n	179 de	30 k
	8	3 7.1 mn	1780 bc	246 ab	68 c-i
lomaine	0	3 97.7 a	39 vwx	67 i-n	79 a-f
Lettuce	2	3 97.7 a 3 1.6 o 3 1.9 o 0 0.6 p	3150 a	267 a	-
•	4	3 1.9 o	<b>4</b> 160 a	222 bc	-
	8	0.6 p	-	-	-
ellow	0	3 99.9 a	34 wxy	47 k-s	47 g-k
Nutsedge	2	3 99.9 a 3 94.1 ab 3 22.2 g-k 3 18.2 ijk	282 r	34 p-s	45 h-k
	4	3 22.2 g-k	719 k-o	80 ij	55 f-k
	8	3 18.2 ijk	962 h-k	67 i-m	101 ab
arley	0	3 98.7 a	41 vw	30 rs	57 f-k
i.u.y	2	3 98.7 a 3 42.8 b-h	1320 c-g	30 rs	59 e-k
	. 4	3 61.3 a-f	1980 Ь	59 j-q	61 e-k
•		3 72.7 a-e	1740 bc	33 q-s	78 a-g

Table 10. Effect of crop species or cultivar and application rate of high Zn sewage sludge on yield, normalized yield, chlorosis severity, and height of second clipping shoots of seven crop species (means of 1 to 8 cultivars per crop).

CROP	N	Rate	Geometric Shoot Yield	Geometric Norm. Yield	Chlorosis Score	Height
			g/pot	%-control	green=1	CTR
Red	16	0	1.55 d-g	93.4 bcd	1.0 g	23.8 h-1
Fescue	18	2	0.41 j	25.4 h-1	2.1 e	20.6 j-m
	20	4	0.46 ij	29.7 g-k	2.7 d	19.6 klm
	13	8	0.14 k	10.1 n	3.7 b	14.9 mn
Merlin	3	0 2	1.11 fgh	78.8 b-e	1.0 g	24.2 h-k
red	3 2 3 3	. 2	0.83 f-j	59.0 b-g	1.0 g	20.7 j-m
fescue	· 3	4	1.06 f-i	75.0 b-f	10 g	25.0 h-k
	3	8	3.43 a-d	244. a	1.3 fg	31.3 efg
Kentucky	22	0	4.78 ab	95.1 bcd	1.0 g	36.5 cde
bluegrass	24	2	1.21 e-h	24.1 j-m	1.8 ef	27.2 f-i
	23	4	1.76 c-f	34.4 f-k	2.0 e	29.0 fgh
	23	8	0.91 f-j	17.9 k-n	2.8 d	21.6 1-1
Tall	23	0 -	6.53 a	99.2 bc	1.0 g	46.3 a
fescue	24	2	2.87 a-d	43.5 d-j	2.9 d	40.6 abc
	24	4	<b>3.77</b> abc	57.2 b-h	3.1 cd	43.0 ab
	24	8	2.69 b-e	40.7 e-j	3.6 bc	38.1 bcd
Perennial	20	0	6.63 a	99.3 bc	1.0 g	32.9 def
ryegrass	21	2	1.63 c-g	24.4 i-m	3.5 bc	26.1 g-j
	21	4	1.15 fgh	17.3 k-n	4.6 a	22.7 1-1
•	21	8	0.71 g-j	10.6 n	4.8 a	21.7 1-1
Canada	2	0	5.24 ab	97.9 bcd	1.0 g	29.0 fgh
bluegrass		2	2.94 a-d	54.8 b-i	2.8 d	24.7 h-k
-	3 3 2	4	0.58 hij	10.9 mn	4.0 b	18.3 lm
	2	8	0.08 k	1.4 o	5.0 a	12.0 n
Cyperus	3	0	3.45 a-d	99.7 bc	. •	•
• •	3	2	4.26 abc	123. ab		•
	3 3 3 3	4	1.62 d-g	46.9 c-j	•	• *
•	3	8	0.44 j	12.9 lmn	•	•

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio t test.

Table 11. Effect of crop species and cultivar, and application rate of high Zn sewage sludge on normalized yield and Zn, Mn, and Fe concentrations in second clipping shoots of seven crop species (means of 1 to 8 cultivars per crop).

Crop .	N	Rat	e Geome Norm.	etric Yield		netric ot Zn	Shoo Mn	t		Shoo Fe	t
<del> </del>			%-cor	ntrol		*****	mg/k	g DW			
Red Fescue	16 18 20 13	2 4	93.4 25.4 29.7 10.1	h-1 g-k	64 822 1340 1800	e-h bcd	64.1 96.2 153. 147.	f-j c-f a ab	•	69.4 86.0 85.5 93.3	def cde cde cde
Merlin red fescue	3 2 3 3	0 2 4 8	78.8 59.0 75.0 244.	b-g	34 211 364 444	k j	79.2 43.5 86.2 87.6	d-i jk c-h c-g		295. 198. 89.1 65.1	a b cde def
Kentucky bluegrass	22 24 23 23	2 4	95.1 24.1 34.4 17.9	j-m f-k	47 583 960 1620	d-g	49.9 48.7 74.7 106.	ijk ijk d-j cd		62.3 84.0 75.3 70.2	def c-f c-f def
Tall fescue	23 24 24 24	2 4	99.2 43.5 57.2 40.7	d-j b-h	53 749 1080 1660	c-f	53.0 49.3 60.7 79.9	h-k ijk g-j c-i		62.3 58.7 55.8 55.2	def def def def
Perennial ryegrass	20 21 21 21	2 4	99.3 24.4 17.3 10.6	i-m k-n	72 947 1350 1890	d-g bcd	58.8 54.1 76.5 101.	g-j g-j d- cde	1	114.4 86.6 108. 87.0	cd cde cde cde
Canada bluegrass	2 3 3 2	2	97.9 54.8 10.9 1.4	b-i mn			57.3 61.0 .96.1 162.	g-j g-j c-f a		54.0 44.2 49.0 18.3	def ef def f
Cyperus	3 3 3	4	99.7 123. 46.9 12.9	ab c-j	65 422 647 1120	h	97.4 19.3 69.8 113.	c-f k e-j bc		.67.0 46.4 100. 137.	def ef cde bc

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio t test.

Table 12. Comparison of the effect of crop species interaction with rate of high Zn sludge on normalized yield (% of control within crop) and Shoot Zn in first and second clipping leaves of 7 crops.

		Geo. Mean Nor	malized Yield	Geo Mean	Shoot Zn	Chlorosis	Score
Сгор	Rate	N Clipping 1	Clipping 2	Clipping 1	Clipping 2	Clipping 1	Clipping 2
		%-Control	%-control	mg/kg	DW	Gre	en=1
Red Fescue	0 2		93.4 bcd	46 W	64 lm	1.0 k	1.0 g
.•		0 33.7 e-j	25.4 h-l	662 m-o	822 e-h	1.2 jk	2.1 e
	4 2		29.7 g-k	942 h-l	1340 bcd	1.6 <sub>-</sub> gh	
	8 2	0 7.5 lm	10.1 n	1440 cd	1800 ab	2.7 d	3.7 b
Merlin		3 99.5 a	78.8 b-e	30 xy	34 n	· 1.0 k	1.0 g
red fescue		3 67.9 a-e	59.0 b-g	88 t	211 k .	1.2 fjk	1.0 g
	•	3 100.6 a	75.0 b-f	165 s	364 j	1.2 ijk	10 g
• .	8	3 83.8 e-d	244.	318 gr	444 13	1.3 h-k	, 1.3 fg
Kentucky	0 2	2 77.1 ab	95.1 bcd	42 w	47 mm	1.0 k .	1.0 g
Bluegrass	2 2	2 40.6 abc	24.1 j-m	415 pq	583 hi	1.2 ijk	1.8 ef
_	4 2	3 39.1 a-e	34.4 f-k	621 a-o	960 d-a	1.5 ghi	2.0 e
	8 2	3 16.6 a-f	17.9 k-n	1020 f-j	1620 ab	2.2 e	2.8 d
Tell	0 2	4 94.0 a-d	99.2 bc	49 UV	53 lm	1.0 k	1.0 g
Fescue ·	2 2	4 90.5 c-1	43.5 d-j	821 i-m	749 gh	1.4 g-j	2.9 d
	4 2	4 70.5 d-1	57.2 b-h	1070 e-i	1080 c-f	1.7 fgh	3.1 cd
	8 2	4 53.3 Jkl	40.7 e-j	1370 c-f	1660 ab	2.0 e	3.6 bc
Perennial	0 2	1 89.5 abc	99.3 bc	. 48 uv	72 l	1.0 k	1.0 g
Ryegrass	2 2	1 67.6 a-e	24.4 i-m	768 j-n	947 d-g	1.7 fg	3.5 bc
	4 2	1 43.0 b-h	17.3 k-n	1010 g-j	1350 bcd	2.1 e	4.6
	8 2	1 27.1 f-i	10.6 n	1425 cde	1890 ab	2.9 d	4.8 a
Canada	0	3 98.4 a	97.9 bcd	42 W	61 lm	1.0 k	1.0 g
Bluegrass	2	3 46.0 a-g	54.8 b-i	573 no	765 fgh	1.7 fgh	2.8 d
	4	3 27.6 f-k	10.9 mm	915 h-l	1380 bc	2.0 ef	4.0 b
•	8	3 3.5 no	1.4 0	1380 cde	2100 a	3.3 c	5.0 a
Yellow	0	3 99.9 a	99.7 bc	34 wxy	65 lm	1.0 k	•
Nutsedge	2	3 94.1 ab	123. ab	282 r	422 1]	1.2 ijk	•
•		3 22.2 g-k	46.9 c-j	719 k-o	647 h	4.0 b	•
•	8	3 18.2 ijk	12.9 lmn	962 h-k	1120 cde	4.0 b	•

Table 13. Variation among cultivars of red fescue in yield reduction, shoot Zn concentration, and chlorosis severity during first growth period on pH 5.5 Sassafras sandy loam amended with four rates of high Zn sewage sludge.

Cultivar	Rate	Geo. Mean Yield	Geo. Normalized Yield	Geo. Mean Shoot Zn	Chlorosis Score
		g/pot	%-Control	mg/kg DW	Green=1
Longfellow	0	3.42 abc	99.6 abc	43 no	1.0 e
	2	2.06 a-f	59.9 a-f	688 hi	1.2 e
	4	0.67 g-j	19.5 g-k	1160 de	2.0 c
	8	0.09 m	2.7 m	1800 ab	3.0 a
Boreal	0	0.94 f-i	73.3 a-f	38 nop	1.0 e
	2	0.13 lm	9.8 jkl	694 ghi	1.0 e
	4	0.19 klm	14.7 g-l	692 hi	1.3 de
	8	0.09 m	7.0 klm	1050 def	3.0 a
Ruby	0	2.99 a-e	97.5 abc	40 nop	1.0 e
	2	3.47 abc	113. a	469 j	1.2 e
	4.	1.08 e-h	35.3 b-h	851 e-h	1.3 de
	8	0.99 f-i	32.1 d-h	991 d-g	2.7 ab
Common Creeping	0 2 4 8	1.09 e-h 0.31 jkl 0.35 i-l 0.08 m	95.1 a-d 26.7 f-j 30.1 e-i 6.6 klm	83 m 643 hij 802 f-i 1170 de	1.0 e 1.2 de 1.3 de 2.5 b
Jamestown	0	3.27 a-d	98.3 abc	37 op	1.0 e
	2	0.81 f-j	24.4 f-j	766 f-i	1.0 e
	4	0.20 klm	6.1 lm	1060 def	1.8 c
	8	0.09 m	2.7 m	1724 abc	2.7 ab
Penn1 awn	0	3.24 a-d	91.1 a-d	38 nop	1.0 e
	2	1.13 d-h	31.8 d-h	595 ij	1.2 e
	4	0.68 g-j	19.2 g-k	836 e-i	1.8 c
	8	0.36 1-1	10.1 i-1	1250 cd	2.5 b
Wintergreen	0	4.17 ab	99.5 abc	53 n	1.0 e
	2	1.39 c-h	33.1 c-h	840 e-i	1.3 de
	4	1.53 b-g	36.6 b-g	1350 bcd	1.7 cd
	8	0.50 h-k	12.0 h-1	2440 a	2.7 ab
Merlin	0	4.48 ab	99.5 abc	30 p	1.0 e
	2	3.05 a-e	67.9 a-f	88 m	1.2 e
	4	4.53 a	101. ab	165 l	1.2 e
	8	3.77 abc	83.8 a-e	318 k	1.3 de

Means in a column followed by the same letter are not significantly different (P < 0.05) according to the Waller-Duncan K-ratio t test.

Table 14. Variation among cultivars of Kentucky bluegrass in yield reduction, shoot Zn concentration, and chlorosis severity during first growth period on pH 5.5 Sassafras sandy loam amended with four rates of high Zn sewage sludge.

		· · · · · · · · · · · · · · · · · · ·		<u> </u>	
Cultivar	Rate	Geo. Mean Yield	Geo. Normalized Yield	Geo. Mean Shoot Zn	Chlorosis Score
		g/pot	%-Control	mg/kg DW	Green=1
Parade -	0	6.25 a	98.2 a-e	. 37 o-r	1.0 g
	2 .	1.82 b-i	28.6 f-m	513 ijk	1.2 efg
•	4	0.96 f-k	15.1 lm	533 1Jk	1.3 d-g
•	8	0.67 ijk	10.6 mno	1140 ab	2.5 a
Rugby .	0	0.95 f-k	32.5 e-m	34 pgr	1.0 g
	2	0.73 g-k	24.8 i-m	395 Jk1	1.0 g
•	4	2.27 å-g	77.6 a-1	689 e-h	1.3 ď-g
•	8	1.16 e-k	39.7 c-1	1100 abc	1.8 bc
Victa		2.51 a-f	72.5 a-j	84 n	1.0 g
•	0 2 4	4.21 abc	122. abc	299 ml	1.2 fg
	4	4.58 abc	132. ab	419 jk1	1.3 d-g
	8	2.18 a-h	62.9 a-k	774 d-g	1.8 bc
Merit	0	3.96 a-d	93.5 a-f	33 qr	1.0 g
	0	3.57 a-e	84.4 a-h	277 m	1.2 fg
	4	4.64 abc		409 jkl	1.5 c-f
	8	2.41 a-f	57.0 a-k	763 d-g	1.8 bc
Merion	0	2.99 a-f	94.4 a-f	47 op	1.0 g
	2	0.71 g-k	22.4 j-m	381 klm	1.0 g
· .	4	0.38 klm	11.9 mm	780 c-g	1.5 c-f
	8	0.13 mm	4.2 no	1040 a-d	2.0 b
South Dakota	0	1.46 c-j	57.0 a-k	44 opq	1.0 g
"Common"	2	0.69 h-k	26.8 g-m	642 hij	1.7 b-e
	4	0.51 jkl	19.8 klm	924 с-е	1.8 bc
	8	0.09 n	3.3 0	1422 a	2.7 a
Mystic	Ŏ	4.99 ab	87.8 a-g	51 o	1.0 g
	2	2.39 a-f	42.0 b-1	539 hij	1.5 c-f
	4	2.60 a-f	45.7 a-1	756 d-h	2.0 b
	8	1.24 d-j	21.8 klm	1180 ab	2.7 a
Bristol	Ŏ	2.63 a-f	148. a	28 r	1.0 g
<b></b>	2	0.59 1-1	33.1 d-m	429 jk	1.0 g
	4	0.46 jkl	26.1 h-m	651 f-h	1.0 g
	8	0.19 1mn		907 b-f	1.8 bcd
·				<i>307 0 1</i>	

Means in a column followed by the same letter are not significantly different (P < 0.05) according to the Waller-Duncan K-ratio t test.

Table 15. Variation among cultivars of tall fescue in yield reduction, shoot Zn concentration, and chlorosis severity during first growth period on pH 5.5 Sassafras sandy loam amended with four rates of high Zn sewage sludge.

Cultivar	Rate	Geo. Mean Yield	Geo. Normalized Yield	Geo. Mean Shoot Zn	Chlorosis Score
		g/pot	%-Control	mg/kg DW	Green=1
Be1-86-1	0	3.27 abc	95.7 a-e	48 †j	1.0 e
	2	2.31 c-h	67.4 e-i	830 fgh	1.3 de
	4	1.83 h-k	53.5 1jk	1110 b-e	1.5 cd
	8	1.24 lmn	36.1 1	1400 abc	2.0 ab
Be1-86-2	0	2.44 b-h	96.9 a-e	50 ij	1.0 e
	2	1.46 i-m	57.8 ij	937 d-g	1.7 bcd
	4.	1.14 mn	45.2 jkl	1170 bcd	1.8 bc
	8	1.00 n	39.6 kl	1400 abc	2.0 ab
Rebel	0	3.40 ab	90.5 a-f	41 j	1.0 e
		3.20 a-d	85.0 b-h	861 e-h	1.5 cd
	4	2.44 b-h	64.9 f-j	954 d-g	1.8 bc
	8	2.25 d-h	59.7 hij	1120 bcd	2.0 ab
Falcon		. 3.17 a-d	96.5 a-e	42 j	1.0 e
•	2	3.58 a	109. ab	723 ĥ	1.3 de
	4	2.90 a-f	88.3 a-g	1010 def	1.8 bc
	8	2.37 b-h	72.4 c-i	1390 abc	2.0 ab
Bonanza	0	2.97 a-e	90.2 a-f	58 <b>i</b>	1.0 e
	2	3.14 a-d	95.5 a-e	827 fgh	1.3 de
	4 .	2.00 f-j	60.9 g-j	998 def	1.7 bcd
	8	1.47 i-m	44.8 jki	1360 abc	2.0 ab
Kentucky-31		2.67 a-g	95.1 a-e	55 i	1.0 e
	2	2.85 a-f	101. a-d	774 gh	1.3 de
•	4	1.96 g-j	69.7 d-i	1100 Б-е	1.7 bcd
	8	1.43 j-n	51.0 i-1	1340 abc	2.3 a
Houndog	0	3.21 a-d	98.7 a-d	52 <b>i</b> j	1.0 e
,	2	3.30 abc	102. a-d	839 fgh	1.3 de
•	4	3.21 a-d	98.8 a-d	1120 bcd	1.5 cd
	8	2.33 c-h	71.7 d-i	1410 ab	2.0 ab
Brookston	0	1.78 h-1	89.0 a-f	50 ij	1.0 e
	2	2.53 a-h	126. a	789 fgh	1.3 de
	4	2.10 e-i	105. abc	1090 cde	1.5 cd
	8	1.29 k-n	64.4 f-j	1550 a	2.0 ab

Means in a column followed by the same letter are not significantly different (P < 0.05) according to the Waller-Duncan K-ratio t test.

Table 18. Effect of Kentucky bluegrass cultivar and rate of high Zn sewage sludge on yield, symptoms, height, and shoot Zn concentration in the Second Clipping Shoots.

Cultivar	Rate %	Geometric Mean Yield	Geo. Norm. Yield	Chlorosis Score	Shoot Height	Geometric Shoot Zn
	······································	g/pot	%-control	green=1	CTE	mg/kg dry
Parade	0	5.03 abc	99.6 a	1.0 1	41.7 a	62 k
	. 2	0.82 f-i	16.3 c	. 1.5 ghi	27.0 d-h	677 g-j
•	4	1.20 c-i	23.8 abc	1.7 f-1	. 28.7 def	
•	8	1.64 b-i	32.4 abc	2.7 bcd	30.3 c-f	1820 ab
Rugby	0	3.50 a-d	82.8 ab	1.0 i	32.3 b-e	56 kl
•	2	1.40 c-i	33.0 abc	2.5 cde	25.0 e-i	520 ij
	. 4	2.57 a-g	60.7 abc	2.8 bc	28.7 <b>d</b> ef	1150 b-f
	8	1.11 d-i	26.3 abc	2.7 bcd	27.3 d-g	2420 a
Victa	0	7.54 a	98.1 a	1.0 i	35.0 a-d	40 klm
	2	1.40 c-i	.18.2 bc	2.5 cde	27.3 d-g	509 ij
	4	3.48 a-e	45.3 abc	2.0 d-g	30.7 b-f	
	8	1.73 b-1	22.5 abc	2.5 cde	22.3 f-j	1190 b-f
Merit	0	7.39 a	98.3 a	1.0 i	39.0 ab	36 lm
	2	2.88 a-g	38.3 abc	1.7 f-i	32.7 b-e	433 j
	4	3.23 a-f	42.9 abc		32.3 b-e	
	8	1.41 c-i	18.7 bc	3.3 ab	23.3 f-j	1310 b-е
South	0	4.08 a-d	97.4 a	1.0 i	41.3 a	50 klm
Dakota	2	1.51 c-i	35.9 abc	1.2 hi	28.0 d-g	
Common	4	1.23 c-i	29.3 abc	1.8 e-h	27.3 d-g	
	8	0.71 ghi	16.9 c	2.3 c-f	18.3 1jk	1890 ab
Merion	0	•	91.3 a	1.0 i	38.0 abc	50 klm
	2	0.83 e-i	22.6 abc		27.3 d-g	
	4	1.51 c-i	41.1 abc	2.3 c-f		
	8	0.07 j	1.9 d	4.0 a	10.3 k	790 f-i
Mystic	. 0	6.70 ab	97.9 a	1.0 i	41.3 a	52 klm
•	2	1.89 a-h	27.6 abc	1.3 ghi	32.0 b-e	683 g-j
	4	1.91 a-h	28.0 abc	2.0 d-g	32.7 b-e	823 e-i
	8	1.62 b-i	23.7 abc	3.0 bc	25.0 e-i	1840 ab
Bristol	0	3.03 a-f	98.9 a	1.0 i	23.0 f-j	34 m
•	2	0.42 i	13.6 c	1.3 ghi	18.7 h-k	427 j
	4	0.49 hi	15.9 c	1.2 hi	20.0 g-j	993 c-g
•	8	0.55 hi	18.0 bc	2.0 d-g-	16.0 jk	1550 abc

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio T-test.

Table 19. Effect of tall fescue cultivar and rate of high Zn sewage sludge on yield, symptoms, height, and shoot Zn concentration in the Second Clipping Shoots.

Cultivar	Rate %	Geometric Mean Yield	Normalized Yield	Chlorosis Score	Shoot Height	Geometric Shoot Zn
		g/pot	%-control	green=1	CIII	mg/kg dry
Be1-86-1	0 2 4	6.62 a-d 2.62 1-1 2.74 1-1	0.976 a 0.392 e-h 0.399 e-h	1.0 g 2.8 def 3.0 cde	42.7 c-i 37.0 g-j 41.3 c-i	47 mn 751 ijk 965 e-i
	8	2.13 kl	0.348 gh	3.8 a	35.3 hij	
Be1-86-2	0 2 4 8	5.71 a-h 2.71 i-1 4.54 a-i 2.65 i-1	1.00 a 0.472 d-h 0.797 abc 0.467 d-h	1.0 g 3.0 cde 2.3 f 3.2 b-e	42.7 c-i 37.7 e-j 44.7 b-g 35.3 hij	47 mn 916 f-j 1220 c-f 1820 ab
Rebel	0 2 4 8	7.30 a 2.23 jkl 3.62 e-1 3.54 f-1	1.00 a 0.332 h 0.513 d-h 0.497 d-h	1.0 g 3.0 cde 3.3 a-d 3.8 a	47.0 a-d 35.7 g-j 41.0 c-i 38.7 d-j	39 <b>m</b> 786 g-j 1090 d-g 1650 abc
Falcon	0 2 4 8	2.52 jk1	1.00 a 0.361 fgh 0.931 ab 0.492 d-h	2.7 ef	50.0 abc 44.3 b-h 44.0 c-h 41.7 c-i	52 lmn 814 g-j 968 e-i 1750 ab
Bonanza	0 2 4 8	6.56 a-d 3.80 c-k 2.77 i-l 2.64 i-l	1.00 a 0.631 cde 0.473 d-h 0.406 e-h	2.8 def	41.3 c-i 31.3 j 34.0 ij 33.7 ij	541 k 935 f-j
Kentucky -31	0 2 4 8	6.80 abc 2.72 i-l 3.95 b-j 2.15 kl	1.00 a 0.406 e-h 0.605 c-g 0.356 gh	1.0 g 2.8 def 3.3 a-d 3.7 ab	54.0 a 47.7 a-d 53.3 ab 42.7 c-i	72 l 792 g-j 1390 a-d 1880 a
Houndog	0 2 4 8	3.80 c-k	U.0//. DCa	1.0 g 2.8 def 3.3 a-d 3.7 ab	46.3 a-f 46.7 a-e 41.7 c-i 40.0 d-j	55 lm 776 hij 1070 d-h 1820 ab
Brookston	0 2 4 8	6.31 a-f 2.95 i-1 3.49 g-1 2.07 l		1.0 g 3.0 cde 3.5 abc 3.7 ab	44.3 b-h 43.7 c-h	71 1 674 jk 1070 d-h 1650 abc

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio T-test.

Table 20. Effect of cultivar of Perennial Ryegrass and rate of application of high Zn sewage sludge on yield, chlorosis, height, and shoot Zn concentration in the Second Clipping Shoots.

Cultivar	Rate	Geo. Mean Yield	Geo. Norm. Yield	Chlorosis Score	Shoot Height	Geo. Mean Shoot Zn
		g/pot	%-Control	green=1	CIII	mg/kg dry
Manhattan	0	6.42 ab	99.6 a	1.0 g	33.3 abc	64 i
	2	1.79 cde	27.8 bcd	3.5 de	27.7 c-f	914 gh
	4	1.50 c-f	23.3 cde	4.5 ab	26.7 d-h	1250 d-g
	8	0.60 gh	9.3 gh1	4.7 ab	21.7 e-j	1990 abc
Ovation	0	5.87 ab	99.8 a	1.0 g	31.0 bcd	75 <b>1</b>
	2	1.41 def	23.9 b-e	2.5 f	24.7 d-j	818 h
	4	0.56 h	9.6 f-1	4.3 abc	18.3 j	1290 def
	8	0.56 h	9.6 f-i	5.0 a	20.3 hij	1590 b-e
Citation	0	6.17 ab	98.7 a	1.0 g	30.3 bcd	76 i.
•••	2	3.19 bc	51.0 ab	3.3 e	27.0 c-g	979 fgh
	4	1.46 c-f	23.3 b-e	5.0 a	24.7 d-j	1450 cde
	8	0.98 e-h	15.6 d-h	5.0 a	25.7 d-h	1900 abc
Prelude	Ŏ	6.80 ab		1.0 g	31.0 bcd	74 1
	2	1.41 def	20.6 c-f	4.0 b-e	23.7 e-j	966 fgh
•	4	1.24 d-g	18.1 c-g		20.7 g-j	1530 b-e
	8	0.82 e-h	12.0 e-i		23.3 e-j	
Yorktown-I	0 1	7.24 a	98.8 a	1.0 g	38.3 a	81 i
	2	0.88 e-h	12.0 e-i	3.7 cde	24.7 d-j	
	. 4	0.48 h	6.6 1	5.0 a	20.3 hij	1300 def
	- 8	0.60 gh	8.1 hi	5.0 a	20.7 g-j	
Premier	0	7.49 a	98.9 a	1.0 g	35.3 ab	73 i
	2	2.41 cd	31.8 bcd	4.2 bcd	28.0 cde	1010 fgh
	2	1.48 c-f	19.5 c-g	5.0 a	23.3 e-j	1540 b-e
	8	0.79 fgh	10.4 f-i	5.0 a	18.7 ij	2240 a
Pennfine	Ō	6.47 ab	99.9 a	1.0 g	31.0 bcd	59 i
	2	1.27 d-g	19.6 c-g	3.3 e	27.0 c-g	981 fgh
	4	2.50 cd	38.7 bc	3.7 cde	25.0 d-1	1150 efg
	: 8	0.71 fgh	· 10.9 e-i	4.2 bcd	21.3 f-j	2040 ab

Means in a column followed by the same letter are not significantly different at the 5% level according to the Waller-Duncan K-ratio T-test.

Cultivar differences in Zn tolerance and uptake Tables 12-15 and 16-20 report the comparisons of cultivars within each of four plant species used in revegetation for the first and second clippings, respectively. Among red fescue cultivars, over all comparisons, only 'Ruby' showed greater Zn-tolerance than other cultivars excepting 'Merlin'. 'Ruby' had somewhat lower shoot Zn than other red fescue cultivars, excepting 'Merlin'.

Among Kentucky bluegrass cultivars, no cultivar had sufficient resistance to soil Zn to merit further evaluation. Among tall fescue cultivars, one was discarded because of poor germination ('Kenhy'). Based on appearance, yield, and shoot Zn, 'Houndog' was the most Zn resistant, while 'Kentucky-31' was the most sensitive. Nearly all tall fescue cultivars were more Zn resistant than any red fescue cultivars (except 'Merlin') or perennial ryegrass. Among perennial ryegrass cultivars, 'Pennfine' was the most sensitive, while 'Prelude' was more resistant. However, none appeared resistant in comparison with the controls (Figure 1).

Concentrations of Cd. Ni. Cu. and Cd:Zn in second clipping shoots The analyses of second clipping shoots for selected cultivars and crops are shown in Tables 21 (crop means) and 22 (crop rate means). As noted above, because the high Zn sludge was used in the experiment, little change would be expected in plant Cd and other trace elements. The analyses shown in Table 23 generally confirm this expectation. Because the sludge applied very little Cd, and the large additions of Zn would be expected to inhibit Cd uptake by roots, added sludge had little effect of plant Cd. A slight increase is observed at the highest sludge-Zn rate, but these plants were suffering severe Zn-phytotoxicity except for 'Merlin' red fescue. In the same way, Cd:Zn in the shoots fell remarkably with increasing applied Zn. These Cd:Zn and shoot Cd results support the importance placed on Cd:Zn ratio in land-applied organic wastes, because severe Zn-phytotoxicity occurred with low normal plant Cd concentrations. A similar effect of added Zn on shoot Zn with little change in plant Cd was observed by Mortvedt (1985) with industrial by-product Zn fertilizers. If a material is low in Cd:Zn, it will have little effect on shoot Cd even when phytotoxicity results from the added Zn (Logan and Chaney, 1983). Cyperus had the highest overall average shoot Cd concentration, but these levels are of no environmental concern.

Interestingly, shoot Cu increased in cultivars/crops where Zn-phytotoxicity occurred at the higher rates of sludge-Zn application. The addition of sludge-Cu at these sludge application rates was at most 56 mg/kg, a level not expected to increase shoot Cu from 5-10 mg/kg to 20-30 mg/kg as observed. Whether this is due to Zn competition for metal adsorption sites in the soil, or possible due to phytosiderophore secretion by the grasses, is not clear.

Shoot Ni was slightly increased at the higher sludge rates, where severe Zn-phytotoxicity occurred. There is a known interaction between Zn and Ni such that added Ni decreases plant Zn, but it seems likely that these small changes are the result of severe yield reductions, or possibly the somewhat lower soil pH at the higher sludge application rates. The Ni concentrations reached were far below Ni-phytotoxic levels, and no useful interpretation can be made.

Thus, the analyses of other elements were largely as expected for a sludge

with very high Zn concentration but low to normal levels of other elements. Only the shoot Cu results differed from predicted results, but the severe Zn-phytotoxicity precludes clear interpretation of shoot Cu changes. Based on the results from this subset of the whole sample set, no further Cd, Ni, or Cu analyses were made. Shoot Pb concentrations varied from 10-100 mg/kg, far higher than expected for plants grown in a greenhouse in these times of low air dispersion of automotive Pb. However, it had previously been shown by Lagerwerff et al. (1973) that the paint used on the metal work of the glass roof in this greenhouse caused substantial Pb pollution of crops grown in this same greenhouse. There was little pattern of sludge addition rate vs. crop Pb, even though the sludge applied high levels of Pb to the soil. Because of this environmental contamination of all samples, crop Pb results are not reported.

Table 21. Effect of crop species (averaged over four sludge-Zn application rates on concentrations of Zn, Cd, Cu, and Ni, and Cd:Zn ratio in second clipping shoots.

	Geometric Shoot Zn	Geometric Shoot Cd	Geometric Shoot Cd:Zn	Shoot Cu	Geometric Shoot Ni
mg/	kg dry wei	ght	X	-mg/kg d	ry weight-
'Ruby' Red Fescue	584 ab	0.68 a	0.12 ab	22.0 a	7.5 a
'Merlin' Red Fescue	170 d	0.14 c	0.067 cd	11.3 b	2.4 cd
'Merion' K. bluegrass	426 c	0.47 ab	0.11 b	18.6 ab	3.9 b
'Ky-31' Tall Fescue.	621 a	0.29 b	0.052 d	17.9 ab	3.4 bc
'Houndog' Tall Fescue	535 ab	0.28 b	0.052 d	15.2 ab	4.0 b
'Manhattan' P. ryegras	s 673 a	0.29 b	0.047 d	18.8 ab	
'Reubens' C. bluegrass		0.36 b	0.091 bc	15.6 ab	
'Folsom' Cyperus	376 c	0.82 a	0.19 a	14.2 b	1.8 d

Means in a column followed by the same letter are not significantly different  $(P \le 0.05)$  according to the Waller-Duncan K-ratio t test using a factorial interaction model (cultivar rate cultivar\*rate block).

Linear regression of shoot Zn on soil Zn Linear regression was used to evaluate the relationship of plant Zn and soil Zn. Although low sludge Zn concentrations produce plateau responses (Corey et al., 1987), high sludge Zn concentrations should produce more nearly linear response. However, at high plant Zn levels, plant uptake slope generally declines.

The results of this analysis are shown in Table 23. Both the original and log transformed data were tested. For the log model, 0 sludge-Zn rate has no meaning, so 5 mg Zn/kg was used, the DTPA-extractable Zn in the control soil. In the analysis of the normal data, significant regressions were obtained for all but 'Merion' Kentucky bluegrass. The slope of 'Merlin' Zn uptake was about 25% that of the other crops, and Cyperus was between 'Merlin' and the other crops.

The log transformed data gave more significant regressions, but it must be recognized that with data covering such a range, log transformation is necessary

Table 22. Effect of crop species and sludge-Zn application rate on concentrations of Zn, Cd, Cu, and Ni, and Cd:Zn ratio in second clipping shoots.

Cultivar	Rate	Geometric Shoot Zn	Geometric Shoot Cd	Geometric Shoot Cd:Zn	Shoot Cu	Geometric Shoot Ni
		mg/kg dry	weight	*	-mg/kg dry	/ weight-
'Ruby' Red Fescue	0 2 4 8	45 lm 679 efg 1040 b 1560 ab	0.43 a-f 1.21 ab 0.57 a-e 0.63 a-e	0.94 ab 0.18 f-j 0.055 k-o 0.040 m-p	8.1 ghi 20.0 a-g 23.4 a-f 31.9 a	4.2 a-g 9.5 a 8.2 ab 9.0 ab
'Merlin' Red Fescue	0 2 4 8	34 m 211 j 364 i 444 hgi	0.31 c-f 0.51 a-f 0.02 g 0.16 f	0.71 abc 0.24 d-h 0.004 q 0.036 m-p	8.2 ghi 11.3 f-i 12.6 e-i 13.4 d-i	2.7 d-i 1.4 ij 2.6 e-i 2.9 c-i
'Merion' Kentucky bluegrass	0 2 4 8	50 klm 879 c 1410 abc 790 def	0.29 c-f 0.66 a-e 0.46 a-f 0.70 a-e	0.58 a-d 0.075 j-n 0.032 nop 0.088 i-m	7.0 hi 18.1 b-h 31.4 a 16.7 c-i	1.9 f-j 4.0 a-g 7.7 ab
'Kentucky -31' Tall Fescue	0 2 4 8	72 kl 792 def 1390 abc 1880 a	0.23 ef 0.33 c-f 0.30 c-f 0.38 a-f	0.30 c-g 0.041 1-p 0.022 op 0.020 p	4.8 i 14.0 d-i 20.4 a-g 25.1 a-e	1.4 ij 3.7 b-h 4.9 a-e 6.4 a-d
'Houndog' Tall Fescue	0 2 4 8	55 kl 776 def 1060 b 1820 a	0.25 def 0.25 def 0.29 c-f 0.36 b-f	0.45 b-f 0.032 nop 0.028 op 0.020 p	5.8 hi 15.3 c-i 23.2 a-f 30.9 ab	2.0 f-j 4.2 a-g 4.5 a-f 6.9 abc
'Manhattar Perennial Ryegrass		76 k 979 b 1450 ab 1900 a	0.16 f 0.27 c-f 0.45 a-f 0.43 a-f	0.20 e-i 0.028 op 0.033 nop 0.023 op	7.1 hi 14.3 c-i 26.1 a-d 26.9 abc	2.2 e-j 4.5 a-f 7.3 ab 7.1 ab
'Reubens' Canada bluegrass	0 2 4 8	61 kl 765 def 1380 abc 2100 a	0.30 c-f 0.27 c-f 0.60 a-e	0.50 a-e 0.035 m-p 0.044 l-p	5.7 hi 18.3 b-h 23.0 a-f	1.3 ij 4.6 a-f 7.1 ab
'Folsom' Cyperus	0 2 4 8	65 kl 422 hi 648 fgh 1120 bcd	0.82 a-d 0.46 a-f 0.89 abc 1.34 a	1.29 a 0.11 h-1 0.14 g-k 0.12 g-k	5.8 hi 6.8 hi 17.8 c-h 23.7 a-f	1.6 hij 1.8 g-j 1.0 j 4.2 a-g

Means in a column followed by the same letter are not significantly different (P  $\leq$  0.05) according to the Waller-Duncan K-ratio t test.

for valid results. In this regression, 'Merlin' had somewhat lower slope and intercept than other crops, and again Cyperus was in the middle of the group of cultivars tested. These regression models can be used to relate the metal uptake of one cultivar to another. In general, the relationships made quantitative in these models is well demonstrated in the Tables of plant responses and composition above.

These regression results strongly support use of Cyperus as a bioassay plant and allow one to relate the results to other tested plants. Another examples of relating relative metal uptake among crops species and cultivars is reported by Carlton-Smith and Davis (1983). This approach has been very important in assessing potential food-chain transfer of sludge-applied Cd, and relative Cd uptake compared to lettuce is summed to represent the whole human diet from garden foods (Chaney et al., 1987).

Table 23. Linear regression models relating soil Zn and shoot Zn of selected cultivars for the second clipping. Both shoot Zn = soil Zn and log(shoot Zn) = log(soil Zn) models were tested.

Cultivar	Intercept	Slope	Adj. R <sup>2</sup>	
Shoot Zn = Soil Zn Models				
'Ruby' Red Fescue	<b>368</b> ·	0.418***	84.3	
'Merlin' Red Fescue	- 89	0.116**	70.7	
'Merion' K. bluegrass	932	0.412	7.5	
'Ky-31' Tall Fescue	351	0.509***	87.4	
'Houndog' Tall Fescue	109	0.486***	94.6	
'Manhattan' P. ryegrass	234	0.496	83.7	
'Reubens' C. bluegrass	207	0.630***	87.3	
'Folsom' Cyperus	-207	0.314	82.5	
L(Shoot Zn) = L(Soil Zn)	Models:			· .
'Ruby' Red Fescue	3.10***	0.546***	98.5	
'Merlin' Red Fescue	2.92**	0.391***	82.6	•
'Merion' K. bluegrass	3.40***	0.535***	88.5	
'Ky-31' Tall Fescue	3.52***	0.496***	98.9	
'Houndog' Tall Fescue	3.05***	0.523***	99.1	
'Manhattan' P. ryegrass	3.42***	0.496***	99.3	
'Reubens' C. bluegrass	3.13***	0.522***	91.8	
'Folsom' Cyperus	3.15***	0.409***	95.6	

<sup>\*\*\*</sup> denotes  $P \le 0.001$ , and \*\*,  $P \le 0.01$  for parameters.

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